

the transcriptional control of said promoter; and wherein the polypeptide is obtained from Lactococcus; and operably linked to a restriction site.

e) ~~at least one restriction site allowing the insertion of a nucleotide sequence of interest under the transcriptional control of said promoter, and wherein the expression cassette does not comprise any part of the sequence encoding the L. lactis ZitS protein.~~

13. (Currently Amended) The expression cassette of claim 12, wherein the p<sub>Zn</sub> ZitR binding site comprises the following sequence:

AAAAATAANGNNNNNNNTTGACATTATTTNNNNNNNTATAT

(SEQ ID NO: 2)

14. (Currently Amended) The expression cassette of claim 13, wherein the p<sub>Zn</sub> promoter ZitR binding site comprises a sequence selected from the group consisting of:

AAAAATAACGTTAACTGGTTGACATTATTTCTTGCTATATAATTAACCATA

(SEQ ID NO: 4); and

AAAAATAACGTTAACTGGTTGACATTATTTCTTGCTATATAATTAACCAGTA

(SEQ ID NO: 5).

15. (Cancelled)

16. (Previously Presented) The expression cassette of claim 12, further comprising a nucleotide sequence encoding an extracellular targeting peptide, operably linked to at least one restriction site for cloning of a nucleotide sequence as a translational fusion with said targeting peptide, wherein the targeting peptide and the at least one restriction site are under the transcriptional control of the p<sub>Zn</sub> promoter.

17. (Currently amended) The expression cassette of claim 16, wherein said extracellular targeting peptide is a signal peptide of sequence comprises the sequence of:

MKKINLALLTLATLMGVSVTVA (SEQ ID NO: 6).

18. (Currently amended) The expression cassette of claim 12 further comprising a nucleotide sequence under the transcriptional control of the p<sub>Zn</sub> promoter fused to a reporter gene, wherein the expression cassette does not comprise any part of the sequence encoding the L. lactis ZitS protein, fused to a reporter gene.

19. (Previously presented) A recombinant vector comprising the expression cassette as claimed in Claim 12 of Claim 12.

20. (Previously presented) A gram-positive bacterium transformed with the expression cassette as claimed in Claim 12 of Claim 12.

21. (Previously Presented) The bacterium of Claim 20, which is a lactic acid bacterium.

22. (Withdrawn) A method of producing a protein in a gram-positive bacterium, which comprises culturing a gram-positive bacterium transformed with at least one expression cassette of Claim 12.

23. (Withdrawn) The method of Claim 22, wherein the grampositive bacterium is a lactic acid bacteria.

24. (Withdrawn) The method of Claim 22, wherein the lactic acid bacteria is selected from the group consisting of lactococci, lactobacilli and streptococci.

25. (Withdrawn) A method of producing a protein in a gram-positive bacterium, which comprises the steps of:

- a) introducing in said bacterium at least one expression cassette of Claim 12, comprising a sequence encoding said protein;
- b) culturing said bacterium in a medium comprising an amount of Zn<sup>+2</sup> that is sufficient to repress the expression of the protein;
- c) inducing the production of said protein by Zn<sup>+2</sup> depletion of said medium; and
- d) recovering the protein produced.

26. (Withdrawn) The method of Claim 25, wherein the Zn<sup>+2</sup> depletion of the medium is effected by adding a divalent cation-chelating compound to the medium.

27. (Withdrawn) The method of Claim 25, wherein the Zn<sup>+2</sup> depletion of the medium is effected by culturing the bacterium until depletion of the Zn<sup>+2</sup> occurs in the medium.

28. (Withdrawn) A method of controlling expression of a promoter of the ZitRSQP operon in a bacterium, which comprises varying concentration of Zn<sup>+2</sup> in a medium containing the bacterium.

29. (Withdrawn) The method of Claim 28, wherein the increasing the Zn<sup>+2</sup> concentration represses expression of the promoter.

30. (Withdrawn) The method of Claim 28, wherein decreasing the Zn<sup>+2</sup> concentration promotes expression of the promoter.

31. (Cancelled)

32. (Previously presented) The expression cassette of claim 31, wherein the sequence encoding the polypeptide has at least 95% identity with the *Lactococcus lactis* ZitR protein.

33. (Cancelled).

34. (Cancelled)

35. (Previously presented) The expression cassette of claim 12, wherein the sequence encoding the polypeptide of b ) has at least 85% identity with GenBank AAK06214 the sequence deposited under accession number AAK06214.

36. (Previously presented) An expression cassette, comprising:

a bacterial promoter  $p_{zn}$  comprising a binding site for the *Lactococcus lactis*, ZitR protein, which site comprises the following sequence:

AAAAATAANGTNNNNNNNTTGACATTATTTT

(SEQ ID NO.: 1)

in which TTGACA is the -35 box of said promoter, and N represents A, C, G or T; and

b) at least one restriction site allowing the insertion of a nucleotide sequence under the transcriptional control of said promoter, and wherein the expression cassette does not comprise any part of the sequence encoding the *L. lactis* ZitS protein.